a23

--44. (amended) Use of a variant of GH1 or GH variant according to claim 12, in the preparation of a medicament, diagnostics composition or kit, or detection kit.-

R E M A R K S

Attached hereto is a marked-up version of the changes made to the claims by the current amendment. The attached page is captioned "VERSION WITH MARKINGS TO SHOW CHANGES MADE".

Respectfully submitted,

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VERSION WITH MARKINGS TO SHOW CHANGES MADE

4. A method according to ~~any preceding claim,~~ claim 1, wherein the individual exhibits normal results in a standard growth hormone function test.

5. A method according to ~~any preceding claim,~~ claim 1, wherein the detection method comprises any sequencing method for determining the sequence of the *GH1* gene of an individual.

6. A method according to ~~any preceding claim,~~ claim 1, wherein the detection method comprises PCR amplification of the *GH1* gene of the individual using (a) a *GH1* gene-specific fragment, being a fragment unique to the *GH1* gene whose sequence is not found in the four other paralogous (non-*GH1*) genes in the GH cluster, and (b) one or more *GH1* gene-specific primers which cannot bind to the homologous flanking regions in the four other paralogous (non-*GH1*) genes in the GH cluster.

7. A method according to ~~any preceding claim,~~ claim 1, wherein the detection method comprises PCR amplification of the entire *GH1* gene of the individual and nested PCR of overlapping constituent fragments of the *GH1* gene of the individual.

8. A method according to ~~any preceding claim,~~ claim 1, wherein the detection method comprises PCR amplification of all or a fragment of genomic DNA spanning the Locus Control Region of the *GH1* gene.

9. A method according to ~~any preceding claim,~~ claim 1, wherein the detection method comprises mutational screening of all or a fragment of the individual's *GH1* gene by DHPLC.

11. A detection method according to ~~any preceding claim,~~ claim 10, which detection method further comprises the use of one or more primer(s) selected from:
CTC CGC GTT CAG GTT GGC (GH1DF);
AGG TGA GCT GTC CAC AGG (GH1DR);
GGG CAA CAG TGG GAG AGA AG (GH2DF);
CCT CCA GGG ACC AGG AGC (GH2DR);
CAT GTA AGC CCA GTA TTT GGC C (GH3DF);
CTG AGC TCC TTA GTC TCC TCC TCT (GH3DR);
GAC TTT CCC CCG CTG GGA AA (GH4DF);
GGA GAA GGC ATC CAC TCA CGG (GH4DR);
TCA GAG TCT ATT CCG ACA CCC (GH5DF);
GTG TTT CTC TAA CAC AGC TCT C (GH5DR);
TCC CCA ATC CTG GAG CCC CAC TGA (GH6DF)
CGT AGT TCT TGA GTA GTG CGT CAT CG (GH6DR);
TTC AAG CAG ACC TAC AGC AAG TTC G (GHD7F);
CTT GGT TCC CGA ATA GAC CCC G (GH7DR);
GTGCCCCAAGCCTTTCCC (LCR15: 1159-1177);
TGTCAGATGTTCAGTTCATGG (LCR13: 1391-1412);
CCTCAAGCTGACCTCAGG (LCR25: 1346-1363);
GATCTTGGCCTAGGCCTCG (LCR23: 1584-1602);
LCR 5A (5′ CCAAGTACCTCAGATGCAAGG 3′);
LCR 3.0 (5′ CCTTAGATCTTGGCCTAGGCC 3′);
LCR 5.0 (5′ CCTGTCACCTGAGGATGGG 3′);
LCR 3.1 (5′ TGTGTTGCCTGGACCCTG 3′);
LCR 3.2 (5′ CAGGAGGCCTCACAAGCC 3′);
LCR 3.3 (5′ ATGCATCAGGGCAATCGC 3′);
GH1G5 (5′ GGTACCATGGCTACAGGTAAGCGCC 3′);
GH1G3 (5′ CTCGAGCTAGAAGCCACAGCTGCCC 3′);
BGH3 (5′ TAGAAGGCACAGTCGAGG 3′);
GH1R5 (5′ ATGGCTACAGGCTCCCGG 3′); and
GH1R3 (5′ CTAGAAGCCACAGCTGCCC 3′).

12. A variant of *GH1*, which differs from *GH1* and is detected by or is detectable by a method according to~~any preceding~~ claim 1 but was not detected by methods used

hitherto, such as those reliant on patient selection criteria based primarily on absolute height.

14. A variant of *GH1* according to ~~any preceding~~ claim 12 comprising a missense mutation.

15. A variant of *GH1* according to ~~any preceding~~ claim 12 comprising a silent mutation which affects the activity of the signal peptide.

17. A protein or amino acid sequence encoded by a variant of *GH1* according to any of ~~claims 12 to 16.~~ claim 12.

21. A screening method for screening an individual suspected of GH dysfunction, which screening method comprises the steps of:
(a) obtaining a test sample comprising a nucleotide sequence of the human *GH1* gene from the individual; and
(b) comparing a region of the sequence obtained from the test sample with the corresponding region of a predetermined sequence
wherein the predetermined sequence is selected from a variant of *GH1* according to ~~any of claims 12 to 16~~ 12.

24. A screening method according to ~~any one of claims 21 to 23,~~ claim 21, comprising:
(a) obtaining a first test sample from an individual; and
(b) comparing the *GH1* gene or *GH1* transcript, or fragment therefrom (*eg* cDNA), in the first test sample to the corresponding gene, transcript or fragment of a *GH1* variant

obtainable from a second test sample derived from an individual exhibiting the following criterion:

(i) growth failure defined as a growth pattern [delineated by a series of height measurements; Brook CDG (Ed) Clinical Paediatric Endocrinology 3rd Ed, Chapter 9, p141 (1995, Blackwell Science)] which, when plotted on a standard height chart [Tanner *et al* Arch. Dis. Child 45 755-762 (1970)], predicts an adult height for the individual which is outside the individual's estimated target adult height range, the estimate being based upon the heights of the individual's parents.

26. A screening method according to ~~any of claims~~ claim 21~~to 25~~ in which simultaneous screens are used either for multiple known mutations or for all possible mutations by hybridization of a labelled sample of DNA (cDNA or genomic DNA derived from the individual) to micro-arrays of mutation-specific oligonucleotide probes immobilised on a solid support.

28. A kit suitable for use in carrying out a screening method according to~~any of claims~~ claim 21~~ to 27~~, which kit comprises:

(a) an oligonucleotide having a nucleic acid sequence corresponding to a region of a *GH1* variant, which region

incorporates at least one variation from the corresponding wild-type hGH gene sequence; and/or

(b) an oligonucleotide having a nucleic acid sequence corresponding to the wild-type hGH gene sequence in the region specified in (a); and, optionally,

(c) one or more reagents suitable for carrying out PCR for amplifying desired regions of the individual's DNA.

30. A kit according to claim 28 ~~or claim 29~~, wherein kit component (a) comprises a plurality of said oligonucleotides immobilised on a solid support.

31. A kit suitable for use in carrying out a detection method in which the variant is at least one of the variants claimed in ~~claims 12 to 16~~ claim 12.

32. A screening method for screening an individual suspected of GH dysfunction, which screening method comprises the steps of:

(a) obtaining a test sample comprising an amino acid sequence encoded by the human *GH1* gene of the individual; and

(b) analysing the test sample for the presence of a GH variant wherein the GH variant is selected from those according to ~~any one of claims 17 to 20~~ claim 17.

34. An isolated, purified or recombinant nucleic acid sequence selected from:

(a) a sequence comprising a variant of *GH1* according to ~~any of claims 12 to 16 or encoding a GH variant according to any of claims 17 to 20~~ claim 12 or

(b) a sequence substantially homologous to or that hybridises to sequence (a) under stringent conditions; or

(c) a sequence substantially homologous to or that hybridizes under stringent conditions to the sequence (a) or (b) but for the degeneracy of the genetic code; or

(d) an oligonucleotide specific for any of the sequences (a), (b) or (c).

37. A process for preparing a variant of *GH1* according to ~~any of claims 12 to 16~~ claim 12, which process comprises:

(i) culturing a host cell ~~according to claim 36~~; and

(ii) recovering from the culture medium the variant of *GH1* thereby produced.

38. An amino acid sequence encoded or expressed by a sequence, vector, or cell as defined in ~~any of claims~~ claim 34~~to 37~~ in culture medium.

39. A composition comprising a variant of *GH1* or a GH variant according to ~~any of claims 12 to 16 or 17 to 20,~~ claim 12, respectively, in association with a pharmaceutically acceptable carrier therefor.

40. Use of a variant of *GH1* or a GH variant according to ~~any of claims 12 to 16 or 17 to 20,~~ claim 12, respectively, for a therapeutic, diagnostic or detection method.

44. Use of a variant of GH1 or GH variant according to ~~any of claims 12 to 16 or 17 to 20 respectively,~~ claim 12, in the preparation of a medicament, diagnostics composition or kit, or detection kit.